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Review

Pathology, Progression, and Emerging Treatments of Peripheral Artery Disease–Related Limb Ischemia

Jonathan Golledge, MChir, FRACS[1,2,3,*]

[1] Queensland Research Centre for Peripheral Vascular Disease, College of Medicine and Dentistry, James Cook University, Townsville, Queensland, Australia
[2] Department of Vascular and Endovascular Surgery, The Townsville University Hospital, Townsville, Queensland, Australia
[3] Australian Institute of Tropical Health and Medicine, James Cook University, Townsville, Queensland, Australia

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ABSTRACT

Purpose: This narrative review summarizes recent research examining treatment targets for peripheral artery disease (PAD)–related limb ischemia.
Methods: Targeted searches of the PubMed and clinical trial registry databases were performed to identify recent findings from animal models of limb ischemia and clinical studies examining PAD progression and treatment. Ongoing clinical trials testing new treatments for PAD were also reviewed. Relevant full-text articles were retrieved and critically reviewed. Where indicated, data were tabulated and summarized in the text.
Findings: Most people with PAD need treatment to improve their walking and function and limit leg pain. Currently, the available treatments of cilostazol, exercise therapy, and revascularization have several deficiencies, including limited access, poor uptake, limited efficacy, and risk of complications. Severe PAD threatens limb viability and is treated by endovascular or open surgical revascularization but is not always successful in achieving limb salvage. Research is ongoing to develop and test new therapies, including new exercise programs, drugs, stem cell treatments and RNA therapeutics, so that new and adjunctive PAD treatments can be offered. Results from multiple clinical trials are expected within the next 5 years.
Implications: It is envisaged that a range of new therapies for PAD will be available in the future.

Introduction

Atherothrombotic disease of the arteries supplying blood to the lower limbs, referred to as peripheral artery disease (PAD), was estimated to affect approximately 5.6% of people 25 years or older in 2015, representing 237 million people worldwide.[1] The number of men and women with PAD was similar in the 25- to 69-year age group, but for people aged ≥70 years there were many more women than men living with PAD (31.7 million vs 22.5 million).[1] People with PAD have a high risk of myocardial infarction, stroke, and cardiovascular death; therefore, control of modifiable risk factors, including smoking cessation, reducing LDL-C, treating hypertension and diabetes, and administration of antithrombotic drugs, is critical in management.[2] The evidence for and treatment of modifiable risk factors for myocardial infarction, stroke, and cardiovascular death has been previously reviewed extensively.[2–4]

Most people with PAD seek medical treatment because of the functional consequences of limb ischemia.[5] The classic symptom of PAD of cramping pain in the calf muscle on walking relieved by rest, known as intermittent claudication, is associated with impaired measure of health-related quality of life, such as low scores on the 36-item Short Form Health Survey.[3,6] Many people with PAD do not have typical symptoms of intermittent claudication.[7] The Walking and Leg Circulation Study identified 6 groups of symptoms (intermittent claudication, exertional leg pain that does not stop the person from walking, exertional pain not involving the calves that does not resolve within 10 minutes of rest, leg pain present on exertion and rest, sometimes asymptomatic, and always asymptomatic) among a group of 415 people diagnosed with PAD by an ankle-brachial index (ABI) <0.9.[7] Only approximately one-third of patients had classic symptoms of intermittent claudication.[7,8] Previous research suggests that irrespective of symptoms all people with PAD have functional impairment in objective tests.[9] The current treatment options for these functional presentations of PAD include center-based or home exercise therapy or oral cilostazol.[3] Currently, guidelines also indicated that in highly selected patients in whom initial treatment options failed that revascularization by endovascular or open surgery can be considered.[10]

The most concerning presenting symptoms of PAD are ischemic rest pain, ischemic ulceration, or gangrene (Figure 1). Ischemic rest pain is

* Address correspondence to: Jonathan Golledge, MChir, FRACS, Queensland Research Centre for Peripheral Vascular Disease, College of Medicine and Dentistry, James Cook University, Townsville, Queensland, Australia, 4811.
E-mail address: Jonathan.Golledge@jcu.edu.au

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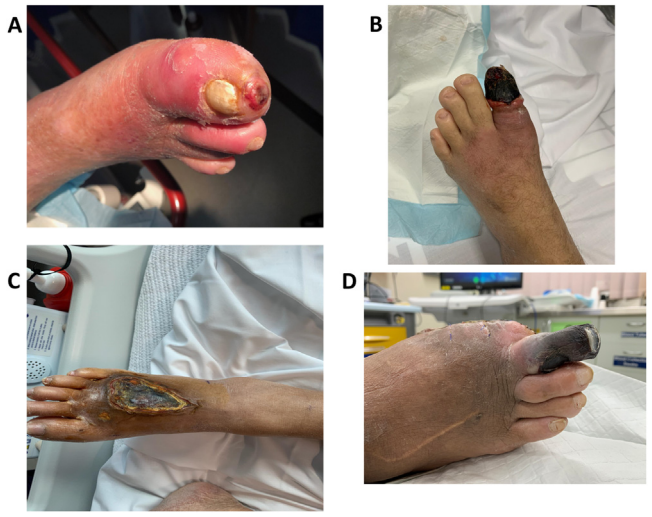

Figure 1. Examples of chronic limb-threatening ischemia. (A) Ischemic ulcer of the right hallux. (B) Gangrene of the left hallux. (C) Extensive gangrene of the right foot. (D) Gangrene of the right second toe after previous hallux amputation.

usually defined by symptoms of severe pain in the foot at rest that keeps the patient awake, requires strong analgesia, is worse when the leg is raised (such as in bed at night) and relieved by dependency, present for at least 2 weeks, and associated with evidence of severe ischemia (ankle Doppler pressure <50 mm Hg or toe pressure <30 mm Hg).[3] When there is objective evidence of PAD, the presentations of ischemic rest pain, foot gangrene, and foot ulceration have been grouped in recent guidelines to be called chronic limb-threatening ischemia (CLTI) as a measure of the substantial risk of major amputation.[3,11] CLTI was previously referred to as critical limb ischemia.[11] The treatment of CLTI focuses on treating any associated infection, analgesia, and endovascular or open revascularization surgery.[11]

Current treatment options for PAD-related limb ischemia have a number of limitations. The only widely approved medication for intermittent claudication, cilostazol, has limited efficacy, is contraindicated in heart failure, and has to be discontinued in up to half of people because of dizziness or palpitations.[12–14] Although there is excellent evidence that exercise therapy improves walking distance in people with intermittent claudication, availability of programs and their uptake is limited.[14,15] There are also limitations of endovascular and open surgical revascularization for treating PAD. Atherosclerosis affects most large and medium arteries, and by the time of symptomatic presentation, most people with PAD have advanced atherothrombosis at multiple sites in their aorta and iliac and infrainguinal arteries. Thus, it is not surprising that after balloon angioplasty, atherectomy, stenting, or bypass revascularization is sometimes not successful in improving function or resolving CLTI.[3,14] Furthermore, the revascularization site can immediately thrombose, leading to acute limb ischemia, which was reported in approximately 8% of patients at 3 years in a recent clinical trial.[16] Repeat interventions and major amputation are still required in up to one-third of patients with CLTI treated by revascularization.[3] People with advanced PAD are also frequently frail and challenging to manage because of complex and competing comorbidities, such as renal failure, cardiac failure, and diabetes.[17]

Thus, a key priority for PAD management is the development of alternative medical treatment options to treat the limb presentations of PAD through improving limb blood supply, minimizing functional decline, and promoting healing of foot wounds. This narrative review summarizes recent research examining new treatment targets in rodent models, human PAD pathology and risk factors, and likely mechanisms of progression of PAD as a basis to inform the development of new therapies. The review focuses mostly on clinical data given the limitations of studying PAD pathology in animal models. Current and emerging treatments for PAD-related limb ischemia are also discussed.

Methods

This narrative review was based on a targeted review of published research, including searches in the PubMed database using the key words *mouse and limb and atherosclerosis, Hind limb ischemia and mouse, "Hind limb ischemia and rat, Hind limb ischemia and rabbit, PAD and histology, PAD and progression, Major adverse limb events, PAD and functional decline, "intermittent claudication and PAD and outcome, major adverse limb events and intermittent claudication, major adverse limb events and CLTI, PAD and walking test, PAD and treatment,* and *PAD and randomized controlled trial*. The ClinicalTrials.gov and the Australia and New Zealand Clinical Trials Registry databases were also searched for ongoing trials in people with PAD. Trials with at least 100 planned participants were focused on. Of particular focus were data on the unique pathology of PAD and targets to limit its progression and the treatment of limb ischemia. Retrieved articles focused mainly on those published very recently in the last 1 to 5 years. Data were handled through tabulation and critical summary.

Animal Models

Types of Models and Translation Challenges

Animal models have high potential for discovery of treatment targets and testing potential new drug therapies. Animal models of PAD focus on inducing hind limb ischemia through artery ligation or excision in mice, rats, rabbits, or pigs.[18] These models have obvious differences from human disease, such as the iatrogenic mechanism of ischemia induction, the focal as opposed to widespread nature of the artery disease, and marked physiologic differences between young, healthy animals and elderly patients with multiple comorbidities.[18] One obvious difference between commonly used animal models of hind limb ischemia and the usual human presentation of PAD is the speed of ischemia onset.[18] Acute hind limb ischemia in most healthy rodent species resolves spontaneously in 1 to 2 weeks. A 2-stage model, in which gradual artery occlusion was caused by placement of 2 ameroid constrictors and at a second operation the femoral artery and associated collaterals were excised, causes more severe and longer-lasting ischemia than femoral artery excision alone.[19] Furthermore this 2-stage ischemia model causes impairment in spontaneous activity and exercise capacity compared with sham controls, unlike the acute ischemia model.[19] The model has thus been suggested as suitable to assess treatments of intermittent claudication. Mice that have undergone the 2-stage model and were repeatedly exercised have functional capacity improvement similar to the response of patients with PAD to exercise therapy.[19] These findings suggest this model maybe useful to test new therapies for PAD-related functional impairment.

Recent Findings Suggesting New Treatment Targets for Limb Ischemia

Table 1 gives examples of interventions recently reported to significantly alter hind limb blood supply in animal hind limb ischemia models. Agents reported to improve hind limb blood supply include stem cells or progenitor cells transfected with microRNA (miR) 221/222[20] or miR-548j-5p[21] to increase angiogenesis-promoting potential; bone marrow–derived mesenchymal stem cells[22]; endothelial progenitor cells combined with an antioxidant[23]; human peripheral blood–derived angiogenic cells[24]; the phosphodiesterase inhibitor sildenafil[25]; viral vectors expressing a flavin adenine dinucleotide-binding protein, which promoted a switch in macrophage phenotype from M1 (proinflammatory) to M2 (anti-inflammatory)[26]; *Bcl2*-associated athanogene 3, which promoted cell viability and necroptosis[27]; hypoxia-inducible factor 1α, which promoted collateral blood vessel expansion (arteriogenesis)[28];

Table 1
Examples of new treatment targets and drug therapies with promising results in animal models.

Species	N (Intervention and Control Group Combined)	Ischemia Induction	Design*			Agent	Proposed Mechanism of Action	Findings
			R	B	S			
Male Sprague Dawley rats	12	Femoral artery ligation	1	1	0	ADSCs transfected with miR-221/222 injected into muscles of ischemic limb†	miR-221/222 proposed endothelial differentiation of ADSC-promoting revascularization	Significantly greater limb perfusion at day 21 after HLI associated with less muscle necrosis and inflammation[20]
Female Sprague Dawley rats	23	Femoral artery excision	0	0	0	Bone marrow–derived mesenchymal stem cells (10^6 IV) 7 days after HLI induction	Increased capillary density and upregulation of VEGF	Significantly greater limb perfusion from days 7 to 21 days after stem cell injection[22]
Sprague Dawley rats with streptozocin-induced diabetes	12	Femoral artery ligation	0	0	0	Endothelial progenitor cells (10^7/100 g) and tempol†	Reduced oxidative stress markers and increased capillary density by activating Wnt signaling	Significantly greater limb perfusion from days 7 to 28 days after stem cell injection[23]
Male Wistar rats	24	Femoral artery ligation	0	0	0	Sildenafil (25 mg/kg/d) started 1 day after HLI induction	Phosphoinositide 3-kinase, Akt, and eNOS activation associated with outward remodeling of small arteries	Significantly greater limb perfusion at day 7 after HLI[25]
Male Sprague Dawley rats	NR	Femoral artery ligation	0	0	0	3×10^5 IU/kg of vitamin D_3 for 3 consecutive days 2 weeks before inducing HLI	Medial calcification induction	Significantly less limb perfusion 21 and 28 days after HLI[32]
Male C57BL/6J mice	12	Femoral artery and collateral excised	0	0	0	Viral vector–expressing period circadian regulator 1 gene injected into muscles of ischemic limb immediately after HLI	Promote macrophage phenotype switch from MI (proinflammatory) to M2 (anti-inflammatory)	Significantly greater perfusion from days 14 to 21 after HLI induction associated with reduced inflammatory markers[26]
Male leptin receptor-deficient mice§	20	Femoral artery excision	0‡	1	0‡	Praliciguat (10 mg/kg/d orally) administered starting 3 days before ischemia induction	Guanylate cyclase stimulator promoting nitric oxide release	More rapid ischemia recovery associated with increased arteriole diameter[29]
Male Albino mice	NR	Femoral artery excision	0	0	0	Fasudil (25 mg/kg/d IP) †	ROCK inhibitor	More recovery of treadmill exercise capacity associated with increase leg muscle antioxidant capacity and reduce muscle apoptosis[30]
Male C57BL/6J mice fed high-fat diet to induce impaired glucose tolerance	NR	Femoral artery excision	1	0	0	Viral vector–expressing *BAG3* injected into leg muscles before HLI induction	*BAG3* promotes cell viability and necroptosis	More rapid ischemia recovery associated with less muscle necrosis[27]
Male C57BL/6J and male and female eNOS-deficient and myoglobin transgenic mice	14	Femoral artery excision	0	0	0	$VEGF_{165}b$ blocking antibody†	Block antiangiogenic effect of $VEGF_{165}b$	Increased VEGFR1-dependent endothelial proliferation and angiogenesis capacity[31]
Male Balb/c nude mice	12	External iliac artery ligation	0	0	0	Human peripheral blood–derived angiogenic cells injected near ligation site at time of HLI induction	Promote angiogenesis	More rapid ischemia recovery associated with less gastrocnemius muscle fibrosis[24]
Male C57BL/6J mice	15	Femoral artery excision	0	0	0	miR-29a viral vector injected into leg muscles 7 days before HLI	Reduced angiogenesis secondary to reduced *ADAM12* expression	Reduced ischemia recovery associated with less leg muscle strength[33]
Male nude mice	12	Femoral artery ligation	0	0	0	miR-548j-5p mimic transfected EPCs injected into leg muscles 2 weeks after HLI induction	Improved capillary density	Improved blood supply to ischemic limb[21]
Male and female New Zealand rabbits		Deep femoral artery ligation and SFAexcised	1	1	0	HIF-1–expressing viral vector injected into leg muscles 7 days after HLI induction	Arteriogenesis	Number of collateral arterioles significantly increased and less signs of muscle damage[28]

ADSC = adipose-derived stem cells; eNOS = endothelial nitric oxide synthase; EPCs = endothelial progenitor cells; HFI-1 = hypoxia-inducible factor 1; HLI = hind-limb ischemia; miR = microRNA; NR = not reported; ROCK = ρ-associated coiled-coil protein kinase; SFA = superficial femoral artery; VEGF = vascular endothelial growth factor; VEGFR = vascular endothelial growth factor receptor.

* Key design features of clinical trials included reported as yes (1) or no (0) for randomized (R) allocation of intervention and control; blinding (B) of outcome assessor and investigators, including placebo-control design; a priori sample size (S) calculation reported testing a clear and supported treatment effect on the primary outcome and met adequately testing the primary hypothesis.

† Timing of agent administration not stated or unclear.

‡ Randomization and sample size calculation were stated to have occurred, but the method reported would not constitute randomization accepted for clinical trials.

§ A small number (11%) were female.

and the guanylate cyclase stimulator praliciguat, which promoteed nitric oxide release.[29] Other interventions reported to be valuable were the ρ-associated coiled-coil protein kinase inhibitor fasudil, which improve exercise capacity recovery,[30] and an antibody blocking the antiangiogenesis isoform of vascular endothelial growth factor, which improved limb capillary density.[31] In contrast, vitamin D_3, which promoted artery calcification,[32] and an miR-29a[33] viral vector both inhibited ischemia recovery. As illustrated in Table 1, the robustness and translation of these findings are unclear based on a number of limitations of most studies, including small sample sizes, absence of methods to avoid confounding required in clinical trials, and design features not relevant to the clinical presentation, such as commencing drug use before ischemia induction or in models in which ischemia is not established but resolving. The tolerability of some of these interventions in humans is also unclear.

Human PAD Pathology

There have been few histologic studies of human PAD pathology. Human atheroma samples have been mainly collected from patients having femoral endarterectomy or major amputation. Narula et al[34] examined the histology of 299 femoral, popliteal, or tibial arteries removed from 75 patients with CLTI and 20 patients without PAD undergoing above- or below-knee amputation. A >70% stenosis as a result of intimal thickening or fibroatheroma or calcified plaque was identified in 69% of the arteries removed from patients with CLTI. Associated thrombosis was significantly more common in arteries that did not have significant atherosclerosis or vessel stenosis than sites with significant atherosclerosis.[34] In a subsequent review article, the authors suggested their findings implicated non–atherosclerosis-stimulated thrombosis as having a more key role in CLTI than acute coronary syndrome.[35] An analysis of femoral atheroma in 51 patients removed at endarterectomy suggested that greater quantities of nodular calcification were present in individuals with less severe limb ischemia.[36] Given the limited and very selected previous investigations, more studies are needed on human PAD pathology.

Because only selected patients have open surgical revascularization and postmortem examinations are rarely performed, imaging studies are important alternative ways to investigate the mechanisms involved in PAD development and progression. Magnetic resonance imaging and intravascular ultrasonography virtual histology has been reported to be representative of conventional histology and more widely applicable.[37,38] In a postmortem study of 12 legs, intravascular ultrasonography had a sensitivity of 85%, specificity of 85%, and an area under the receiver operator curve of 85% in identifying intimal calcification found during conventional histology.[38] An intravascular ultrasonography study of 159 patients undergoing endovascular revascularization found that 28 (18%) had plaque rupture.[39] Currently, magnetic resonance imaging or high-resolution ultrasonography is not widely used to monitor PAD progression. Expansion of studies of this type may provide important insight into new PAD treatment targets.

Human PAD Progression

There is no agreed-on definition of human PAD progression. A variety of approaches have been used to study progression of disease, including imaging follow-up, symptom recording, functional assessment, and incidence of major adverse limb events, and the recent findings of these studies are discussed in this section.[3,40]

Imaging Assessment of PAD Progression

The ABI is a cheap and reliable way of assessing limb blood supply recommended for PAD diagnosis.[10,41] Falsely elevated ABI can occur in patients with diabetes or renal failure but still provides an important measure of increased cardiovascular event risk.[42] Monitoring PAD progression using ABI, however, is not straightforward for a number of reasons. First, hospital populations frequently have revascularization procedures that are expected to influence ABI. Second, small changes in ABI may result from measure error. Third, it is complex to incorporate changes in ABI in both legs. These difficulties may explain why few studies monitoring progression using ABI were identified. The few studies found included a mix of people with and without PAD recruited from the community. A study of 508 people recruited from the vascular laboratory in San Diego, California, reexamined ABI a mean of 5 year later.[43] Approximately 17% of patients had a decrease in ABI of ≥0.15 at follow-up, which was associated with an increased risk of cardiovascular (relative risk [RR] = 2.8; 95% CI, 1.3–6.0) and all-cause (RR = 2.4; 95% CI, 1.2–4.8) mortality.[43] A recent study investigated 681 patients with type 2 diabetes recruited from a tertiary health care facility in Rio de Janeiro, Brazil.[44] Seventy-seven (11.3%) developed an ABI ≥0.9 or had a decrease in ABI of >0.15 when recordings were repeated a mean of 91 months later. Independent risk factors for developing ABI ≥0.9 or a decrease in ABI of >0.15 were longer diabetes duration, male gender, microvascular complications (neuropathy and kidney disease), higher systolic blood pressure, and LDL-C.[44]

Ultrasonography, computed tomography, and magnetic resonance imaging are likely to be more sensitive in detection of progression of peripheral atherosclerosis than ABI, but there is currently no agreed-on definition of what constitutes PAD progression using these modalities. Furthermore, imaging monitoring of people with PAD is not currently indicated in practice guidelines; thus, the population with this type of follow-up is limited and selective. Futures studies need to clarify whether there is value in using such image to monitor people with PAD.

Clinical Assessment of PAD Progression

Clinical progression of PAD is most commonly defined by the incidence of major adverse limb events, typically defined to include amputation at or proximal to the ankle (major amputation) and requirement for surgical or endovascular revascularisation.[11] Table 2 gives the rates of major adverse limb events, major amputation, and revascularization among different populations with PAD reported in publications during the last 3 years.[45-53] The 1-year rates of these events are reported to be highly variable: 4% to 46% for major adverse limb events,[46,48,50] <1% to 41% for major amputation,[45,46] and 4% to 32% for revascularization (Table 2).[46,48] This variation in the incidence of major adverse limb events reported in previous studies likely results for variations in the risk factors in the population studied. Risk factors for increased adverse events identified in recent studies include underrepresented ethnicities, such as Black or Hispanic Americans, younger age, CLTI, smoking, history of cancer, amputation, lower limb revascularisation, ischemic heart disease, dementia or renal failure, severe ischemia (evidenced by low ABI), and increased inflammatory markers, such as high erythrocyte sedimentation rate, distal site of intervention, and revascularization by endovascular as opposed to open surgery (Table 3).[47,49–52,54]

Progression of PAD in people with intermittent claudication has also been monitored from patients' symptoms (eg, development of symptoms and signs of CLTI). A retrospective comparison of patients presenting with intermittent claudication and CLTI proposed a score to predict development of CLTI, which included older age, male gender, diabetes, higher serum concentrations of uric acid and triglyceride, high mean heart rate, and body mass index.[55] The score had an area under the receiver operator characteristic curve of 0.73 for assessing the distinction between intermittent claudication and CLTI using their cross-sectional population.[55] This score needs to be assessed on prospectively followed up cohorts to adequately assess its clinical value. The Examining Use of tiCagreLor In paD (EUCLID) trial examined symptom progression using the Rutherford classification and reported that older age, diabetes, and lower ABI were independently associated with worsening symptoms during 12 months follow-up.[56]

Table 2
Incidence of major adverse limb events.

Design	N	Female, No. (%)	Country/ Ethnicity	IC, No. (%)	CLTI, No. (%)	Initial Management	Follow-up, mo*	Majo Adverse Limb Events, No. (%)	Major Amputation, No. (%)	Lower Limb Revascularization, No. (%)
RCT[45]	200	56 (38)	Australia	200 (100)	0	Exercise advice	24	23 (11.5)	2 (0.5)	23 (11.5)
Kleiss et al[46]	81	28 (34.6)	Netherlands	0	81 (100)	EVT infrapopliteal reintervention	12	46%‡	41%‡	32%‡
Cohort[47]	692	182 (26.3)	Japan	547 (79.0)	145 (21.0)	EVT	40	NR	NR	159 (23.0)
Cohort[48]	500	110 (22)	Japan	500 (100)	0	EVT plus HEP	12	22 (4.4)	1 (0.2)	22 (4.4)
Registry[49]	67,651	23,675 (35)	United States/White	31256 (46.2)	36,395 (53.8)	EVT or open revascularization	8	NR	1827 (2.7)	NR
Registry[49]	15442	7048 (45.6)	United States/Black	5297 (34.3)	10,145 (65.7)	EVT or open revascularization	8	NR	896 (5.8)	NR
Registry[49]	5506	2155 (39.1%)	United States/Hispanic	1796 (32.6)	3710 (67.4)	EVT or open revascularization	8	NR	308 (5.6)	NR
EMR[50]	1209	489 (40.4)	United States/31% Black and 69% White	730 (60.4)	479 (39.6)	Conservative	12	551 (45.6)†	147 (12.2)	285 (23.6)
Registry[51]	2913	721 (24.8)	NR	2913 (100)	0	Infra-inguinal bypass	12	Approximately 15%‡	Approximately 2%‡	NR
Database[52]	247	95 (38.4)	United States/7.5% Black, 29.9% Hispanic, 44.5% White, 13.9% Asian, and 4.2% other	0	247 (100)	EVT or open revascularization	60	Approximately 53%‡	Approximately 23%‡	Approximately 38%‡
Database[53]	6809	2738 (40.2)	90% White, 5.8% Black, and 4.2% other	3334 (49.0)	3475 (51.0)	EVT	24	954 (14.0)	272 (4.0)	682 (10.0)

CLTI = chronic limb-threatening ischemia; EMR = electronic medical record; EVT = endovascular revascularization therapy; HEP = home exercise program; IC = intermittent claudication; NR = not reported; RCT = randomized clinical trial.

* Median, mean, or set follow-up months.

† Combined major adverse cardiovascular events and major adverse limb events.

‡ Only available from survival curves.

Table 3
Risk factors for major amputation and revascularization.

Study	Initial Management	Risks Factors for Different Events (Adjusted Hazard Ratios and 95% CIs)		
		Major Adverse Limb Events	Revascularization	Major Amputation
JPASSION[47]	EVT	NR	CLTI: 1.98 (1.00–3.91), ESR: 1.01 (1.00–1.03), infrainguinal intervention: 3.49 (1.33–9.16)	NR
VQI Registry[49]	EVT or open revascularization	Black Americans: 1.20 (1.10–1.20); Hispanic Americans: 1.20 (1.10–1.30)	NR	Black Americans: 1.50 (1.40–1.70); Hispanic Americans: 1.50 (1.30–1.60)
Duke University Health System Study[50]	Conservative	NR	CLTI: 6.60 (3.97–10.99); age: 0.98 (0.96–1.00); Black Americans: 0.69 (0.49–0.99); history of cancer: 1.52 (1.05–2.18); prior amputation: 0.58 (0.37–0.91); mean ABI: 0.41 (0.22–0.77)	CLTI: 27.29 (9.68–76.93); COPD: 0.52 (0.32–0.87); dementia: 4.43 (1.89–10.36); IHD: 1.97 (1.08–3.58); prior amputation: 2.30 (1.30–4.07)
K-VIS ELLA[54]	EVT	IDDM: 1.39 (1.02–1.89); ESRF: 1.78 (1.24–2.55); past amputation: 1.56 (1.02–2.41); previous EVT: 1.86 (1.21–2.85); CLTI: 1.97 (1.43–2.72)	ESRF: 1.72 (1.07–2.75); previous EVT: 2.33 (1.46–3.74); HbA_{1c} ≥7%: 1.47 (1.01–2.16)	NR
VQI[51]	open revascularization	Current smoking: 1.28 (1.01–1.66)	NR	Black race: 2.82 (1.30–6.17), below-knee bypass grafting: 2.50 (1.21–5.17), prosthetic conduit: 1.97 (1.06–3.67)
Stanford[52]	EVT or open revascularization	Open vs EVT: 0.39 (0.17–0.91)	Open vs EVT: 0.35 (0.13–0.96)	None identified
VSGNE[53]	EVT	Therapeutic anticoagulation: 1.39 (1.09–1.76)	NR	NR

ABI = ankle brachial index; COPD = chronic obstructive pulmonary disease; ESRF = end-stage renal failure; EVT = endovascular revascularization therapy; HbA_{1c} = glycosylated hemoglobin; IDDM = insulin-dependent diabetes; IHD = ischemic heart disease; JPASSION = Japan Peripheral Artery disease: endovaScular revaScularizatION prospective multicenter observational study; K-VIS ELLA = Korean Vascular Intervention Society Endovascular therapy in Lower Limb Artery diseases registry; NR = not reported; VSGNE: Vascular Study Group of New England; VQI = Vascular Quality Initiative.

Table 4
Change of function using 6-minute walking test in people with PAD.

Design	N	Female, No. (%)	Country/Ethnicity	CLTI, No. (%)		Management	Follow-up, mo	Six-Minute Walking Distance, Mean (SD)		
				No	Yes			Baseline	Follow-up	MD
Single arm trial[66]	158	37 (31)	China	138 (87)	20 (13)	EVT with paclitaxel-coated balloon	12	262 (96)	NR	63 (98)
Single arm trial[61]	95	31 (33)	Switzerland	95 (100)	0	Supervised exercise of walking and strengthening	3	418 (399 to 437*)	469 (453–485)	51†
RCT sub-study[60]	45	24 (53)	67% Black			Supervised exercise attendance tertile 1 (11-68%)	6	307 (90)		28 (1.3 to 54*)
	46	19 (41)	57% Black			Supervised exercise attendance tertile 2 (69-<85%)	6	338 (94)		38 (12 to 64*)
	46	20 (43)	48% Black			Supervised exercise attendance tertile 3 (≥85%)	6	361 (99)		57 (30 to 84*)
RCT[63]	54	23 (43)	Sweden	54 (100)	0	Supervised exercise of Nordic walking	12	385 (361 to 411*)	396 (368, 424*)	−12 (−36 to 13*)‡
	56	21 (38)	Sweden	56 (100)	0	Home exercise of Nordic walking	12	387 (362 to 411*)	406 (377, 434*)	
RCT[64]	116	54 (47)	56% Black and 39% White	116 (100)	0	Low intensity home exercise	12	332 (96)	328 (109)	−6 (−22 to 9*)
	124	60 (48)	65% Black and 33% White	124 (100)	0	High intensity home exercise	12	338 (103)	371 (117)	35 (20 to 49*)
	65	32 (49)	54% Black and 43% White	65 (100)	0	Non exercise control	12	328 (91)	318 (99)	−15 (−36 to 6*)
RCT[65]	95	29 (31)	12% Black and 76% White	95 (100)	0	Home walking behavior change counselling	3	353 (87)	381 (88)	22 (1 to 44*)
	95	28 (29)	7% Black and 82% White	95 (100)	0	Usual care	3	370 (78)	372 (77)	9 (−15 to 34*)
RCT[45]	102	29 (28)	Australia	102 (100)	0	Brief behavior counselling	24	372 (93)	364 (109)	2 (−19 to 23*)‡
	98	27 (28)	Australia	98 (100)	0	Attention control	24	373 (102)	360 (104)	
RCT[69]	59	22	73% Black American	59 (100)	0	Telmisartan	6	342 (93)	343 (93)	−17 (−36 to 2*)‡
	55	24	69% Black American	55 (100)	0	Placebo	6	352 (116)	365 (107)	
RCT[67]	18	5	33% African American	18 (100)	0	Heat leg therapy trousers	2	368 (89)	NR	21 (10 to 42§)
	16	3	20% African American	16 (100)	0	Sham therapy	2	396 (66)	NR	−1 (−6 to 14§)

MD = mean difference; NR = not reported; RCT = randomized controlled trial; NS = nonsignificant difference.
* 95% CIs not SD.
† Calculated not reported.
‡ MD in change in walking distance between randomized groups for intervention.
§ Median and 75% CIs.

Monitoring PAD Progression Through Functional Assessment

Limitations in function, particular walking, is an important consequence of PAD, and the functional deficit is strongly correlated with quality-of-life impairment.[6] Decline in function is common among people with PAD, with the **W**alking and Leg Circulation Study reporting that 57 of 277 participants (21%) lost the ability to walk continuously for 6 minutes in 5 years.[57] ABI <0.5 compared with a normal ABI increased the risk of mobility loss by 30-fold.[57] Patients with PAD have evidence of lower limb myopathy identified in imaging and histology studies.[5,58] These findings highlight the importance of effective treatments to maintain activity and function in patients with PAD.

A recent transcriptomic study identified a number of RNAs and pathways differentially expressed in the gastrocnemius muscles of patients with PAD compared with controls, such as upregulation of myosin binding protein H and α-actin cardiac muscle 1 and downregulation of fibroblast growth factor–binding protein 2, suggesting possible targets to reduce functional decline.[59] Table 4 gives the findings of recent cohort studies and randomized controlled trials examining change over time in 6-minute walking distance as a measure of function in people with PAD. Recent studies have reported that supervised exercise improved walking distance by between 28 and 57 m during 6 months, depending on session attendance.[60,61] Structured home exercise programs have also been found in some but not all randomized controlled trials to improve walking distance.[62,63] The benefit likely depends on the frequency and intensity of exercise achieved by the program.[45,60,64] A recent trial reported that a motivational counseling program improved walking distance by 22 m greater than usual care.[65] In contrast, a brief counseling program consisting of only 2 face-to-face and 2 telephone sessions had no effect on 6-minute walking distance compared with attention controls.[45] Although a recent network meta-analysis of randomized controlled trials reported no significant benefit, a recent cohort study found that endovascular revascularization improved walking distance by 63 m at 12 months.[14,66] A recent small randomized controlled trial (n = 34) compared walking in patients randomly allocated to wear trousers that circulated hot water (43°C) compared with cold water (33°C).[67] Participants were asked to apply the therapy for 90 minutes daily. By 8 weeks heat therapy was reported to significantly improve 6-minute walking distance by a mean of 21 m, unlike cold water, albeit nonadherent participants were excluded (Table 4).[67] Prior mice studies suggest that heat therapy upregulated endothelial nitric oxide synthase expression promoting collateral blood flow.[68] Finally,

Table 5
Ongoing randomized controlled trials in people with peripheral artery disease.

Trial Registration	Planned Sample Size	Treatment	Control	Follow-up, mo	Primary Outcome	Secondary Outcomes	Expected Completion
NCT05132439[85]	200	Metformin XR 1000 mg/d	Placebo	6	6MWT	QOL questionnaires, other functional assessment, and blood biomarkers	March 2030
NCT03054519[87]	212	Metformin 2000 mg/d	Placebo	6	6MWT	Treadmill walking, flow-mediated dilatation, WIQ, SF-36, calf muscle biopsy	Sept 2024
ACTRN12618001186246[86]	250	Metformin 1500 mg/d	Placebo	6	6MWT	Physical activity, QOL, ABI, biomarkers	NR
ACTRN12621001383853[92]	180	Footplate muscle stimulation	Sham	5	6MWT	QOL, physical activity, treadmill walking distance, adherence, tissue oxygenation	NR
NCT05465070[93]	106	Heat therapy	Sham	3	6MWT	WIQ, SF-36, SPPB, blood pressure	April 2027
NCT05624125[88]	210	beetroot juice contains 400 mg of nitrate twice daily	Placebo beetroot juice filtered to remove nitrate	4	6MWT	WIQ, calf muscle perfusion, SF-36, SPPB, gastrocnemius nitrate, and mitochondrial oxygen consumption	Dec 2027
NCT04228978[94]	212	Weight loss and home exercise	Exercise alone	12	6MWT	Physical activity, WIQ, patient-reported outcomes of mobility	May 2027
NCT04794530[89]	190	Cocoa flavanols	Placebo	6	6MWT	Gastrocnemius muscle perfusion (MRI), flow-medicated dilatation, physical activity, calf eNOS	Dec 2026
NCT03871075[91]	230	Intermittent pneumatic compression and home exercise or Intermittent pneumatic compression alone	Sham control plus home exercise or sham control alone	6	6MWT	Gastrocnemius muscle perfusion (MRI), flow-medicated dilatation, physical activity	Oct 2024
ACTRN12619000423112[90]	120	Mirabegron	placebo	3	Treadmill maximum walking distance	Physical activity, flow-mediated dilatation, MRA blood flow, endothelial progenitor cell function	NR
NCT05166187[100]	150	BPA as part of discharge workflow	Same care without BPA window during discharge	1.5	Frequency of high intensity statin prescription at discharge and 90 days	Cardiovascular events, including MI, coronary revascularization, stroke, lower limb revascularization, or all-cause mortality	Dec 2023
ACTRN12621001475831[99]	246	Holistic medical management program	Usual care	6	Composite of control of modifiable risk factors	Blood pressure, HbA_{1c}, LDL-C, smoking, physical activity, QOL, 6MW, engagement, biomarkers	NR
NCT04774159[101]	150	Colchicine 0.5 mg	Placebo	12	Mean number of patients recruited per center per month	Proportion of patients randomized after active run-in, loss to follow-up, drug discontinuation and adherence	Sept 2024
NCT04772300[102]	230	Sirolimus-coated balloon catheter	Noncoated balloon catheter	6	Limb salvage and primary patency	Major adverse limb events, revascularization, patency, QOL, mortality, amputation free survival, ABI	July 2027
NCT04982367[103]	166	Sirolimus-coated balloon catheter	Paclitaxel-coated balloon catheter	12	Primary patency	Revascularization, Rutherford class, ABI, tolerability	July 2024
NCT03975946[104]	260	Rheopheresis apheresis technique	Sham	8	Percentage complete wound healing; major amputation	NR	June 2024

ABI = ankle brachial index; BPA = best practice advisory; eNOS = endothelial nitric oxide synthase; HbA_{1c} = glycosylated hemoglobin; MI = myocardial infarction; MRA = magnetic resonance angiography; MRI = magnetic resonance imaging; NR = not reported; QOL = quality of life; SF-36 = 26-item Short Form Health Survey; 6MWT = 6-minute walk test; SPPB = short performance battery; WIQ = walking impairment questionnaire.

a small randomized trial found no benefit of telmisartan on walking distance.[69]

Treatment of Leg Ischemia

Treatments for Intermittent Claudication

The main treatment goal of most patients with intermittent claudication is to improve walking distance and function.[70] Supervised center-based exercise therapy consisting of treadmill-based walking at approximately 50% maximum workload, which induces claudication within 3 to 5 minutes, is the recommended first-line therapy for walking impairment caused by PAD.[71] Sessions of 30 to 50 minutes performed 3 times per week for at least 12 weeks are advised. It is recommended that every few weeks there is progression in training load or time. Maintenance of at least 2 sessions per week is recommended after the 12-week program. For patients unable to attend center-based exercise programs or in countries where supervised exercise programs are not funded, structured home exercise is an alternative, although the most effective program format is less clear.[62,71]

The main issue with exercise programs is their variable uptake by health care systems, clinicians, and patients.[15] A study of 489 people with PAD living in Chicago, Illinois, found that 416 (85%) reported that their physician had never prescribed or recommended supervised

exercise therapy.[72] Most patients indicated willingness to travel 3 times per week to the exercise therapy center, but 60% indicated inability or unwillingness to pay $11 per exercise session copayment. Only 10% of the patients referred for supervised exercise completed the 12-week program. Stated reasons for this included inability to commit the required time and that treadmill exercise was not interesting.

Drug therapies for intermittent claudication are more controversial. Current guidelines recommend the use of cilostazol, but meta-analyses report variable benefits in walking distance.[14,73,74] Cilostazol is also contraindicated in heart failure and has been reported to be discontinued in up to half of patients within 3 months.[13] Pentoxifylline and beraprost have been reported to improve walking distance in some small randomized trials with high risk of bias and are not currently recommended by practice guidelines.[73,75,76]

The role of revascularization in treating intermittent claudication is controversial. Current guidelines indicate that it is reasonable to consider revascularisation for lifestyle-limiting intermittent claudication.[10,77,78] A recent meta-analysis of randomized controlled trials, however, suggest that revascularization treatment alone does not improve walking distance in people with intermittent claudication.[14] There are also concerns about tolerability, with some observational studies finding that people treated invasively have a higher major amputation rate than those managed conservatively during longer-term follow-up.[79] A recent meta-analysis of randomized controlled trials of people with intermittent claudication reported no significant effect of revascularization on major amputation rate.[80] The RR was concerning at 1.69, but the 95% CIs were very wide and crossed 1 (0.54–5.26) and the analysis likely underpowered (n = 1477) to exclude an important increased risk of amputation.[80] Both observational studies and a meta-analysis of randomized controlled trials found that revascularization increases the rate of additional invasive procedures (RR = 4.15; 95% CI, 2.80–6.16).[79,80] These data suggest that the revascularization should be used very selectively in treating intermittent claudication.

Treatments for CLTI

The established treatments for CLTI are endovascular and open surgical revascularisation.[10,11,52,77,81] Techniques for endovascular revascularization have evolved rapidly during the past decade and now include a range of atherectomy devices, drug eluting balloons and stents, along with traditional angioplasty balloons and bare metal stents.[82,83] Most people with CLTI have a range of comorbidities, are elderly, and are frequently frail.[17] Thus, endovascular revascularization has been increasingly considered the first-line treatment for many patients with CLTI. The recent Best Endovascular versus Best Surgical Therapy in Patients with CLTI (BEST-CLI) randomized trial examined whether patients with CLTI were best treated initially with endovascular or open surgical revascularization.[84] The trial included 2 cohorts. Cohort 1 (n = 1434) included patients with great saphenous veins suitable for infrainguinal bypass, whereas cohort 2 (n = 396) included patients with no suitable great saphenous veins who were expected to require a prosthetic conduit for bypass if treated by open surgical revascularization. In cohort 1, major adverse limb events or all-cause mortality was significantly lower after surgical (42.5%) than endovascular (57.1%) revascularization (hazard ratio [HR] = 0.68; 95% CI, 0.59–0.79), as was major amputation (HR = 0.73; 95% CI, 0.54–0.98). In the smaller cohort 2, there was no significant difference in these outcomes between treatments. The risk of reintervention was substantially lower after surgical compared with endovascular revascularization in both cohorts (HR = 0.35; 95% CI, 0.27–0.47 for cohort 1 and HR = 0.47; 95% CI, 0.29–0.76 for cohort 2). The findings suggest that when great saphenous vein is available open surgical revascularization is the ideal treatment for CLTI cause by infrainguinal PAD. Some caveats should be noted, such as the participants in BEST-CLI were relatively young (mean age, <70 years) and thus may have been less frail than those commonly presenting for treatment outside randomized controlled trials. This needs to be considered in generalizing the findings of BEST-CLI.

Future Treatments

A search of the US, Australian, and New Zealand clinical trial registries identified multiple ongoing randomized controlled trials testing new therapies for PAD (Table 5). Most are testing new or repurposed drugs for walking impaired by PAD. Metformin is being tested in 3 trials.[85–87] Other interventions being investigated include beetroot juice, cocoa flavanols, the β_2-agonist mirabegron, intermittent pneumatic compression, heat therapy, weight loss, and neuromuscular stimulatory foot pad.[88–94] These interventions have been supported by prior studies in animal models or pilot human trials. People with PAD have endothelial dysfunction, which limits the ability to produce nitric oxide, a key vasodilator.[95] Increasing evidence suggests that dietary inorganic nitrates, such as provided from concentrated beetroot juice, can improve peripheral perfusion and walking distance in people with PAD. A small clinical trial reported that concentrate beetroot juice increased plasma nitrite concentrations and walking distance in 8 people with PAD.[96] These findings were confirmed in a 24-patient randomized trial in which beetroot juice plus exercise therapy increased 6-minute walking distance by a mean of 53 m compared with 25 m for exercise and placebo.[97] In contrast, a recent study found no effect of dietary inorganic nitrates on walking distance in 18 patients with PAD.[98] An ongoing, larger randomized trial (n = 210) is testing the effect of beetroot juice on walking distance.[88] Other nitric oxide donors are being developed for use in PAD, although it remains to be seen whether they will be effective and well tolerated.[29]

A number of programs aimed at improving the prescription of medical therapy or the control of modifiable risk factors for cardiovascular events are also being trialed.[99,100] A vanguard study is also examining the feasibility of low-dose colchicine as a new treatment to prevent major adverse events in PAD.[101] Finally, a range of interventional therapies are being tested, such as drug-coated balloons and an apheresis method.[102–104] It is hoped that these trials will identify a range of new effective therapies for PAD.

Conclusions

Most people with PAD need treatment to improve their walking and reduce leg pain. Currently, the available treatments of cilostazol, exercise therapy and revascularization have several limitations. Severe PAD threatens major amputation and is treated by endovascular or open surgical revascularization but is not always successful in achieving limb salvage. Research is ongoing to develop and test new therapies, including new exercise programs, drugs, stem cell treatments, and RNA therapeutics to offer new and adjunctive PAD treatments, with results from multiple clinical trials expected with the next 5 years.

Declaration of Competing Interest

None.

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Author Contributions

JG performed all aspects of this study including literature review, data collection, data interpretation and article writing.

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